

## The primary vascular system and medullary bundle structure of *Phytolacca dioica* (Phytolaccaceae)

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### **Abstract:**

*Phytolacca dioica* has a primary vascular system which includes medullary bundles. The primary structure of these bundles is composite, consisting of two to four collateral vascular strands with their phloem poles oriented toward a common center. A cambium is formed between the xylem and phloem of the strands and extends to enclose the phloem of the whole bundle. After a period of cambial activity the medullary bundles become amphivasal. As is typical of species with helical phyllotaxy, the primary vascular system is organized into sympodia. The medullary bundles form the distal portions of the median leaf traces and continue in a medullary position for the number of nodes equal to the denominator of the phyllotactic fraction characterizing a given stem. As a medullary bundle passes out into a leaf, two or three vascular strands pass inward from the vascular cylinder to form a new medullary bundle. The number of medullary bundles in a stem is, thus, maintained. Variations of this pattern occur in the basal regions of juvenile shoots and in the basal and apical regions of adult flowering shoots. The relationship between leaf arrangement and the passing of vascular strand into the pith is discussed and a new classification of vascular systems with medullary bundles is proposed.

Des faisceaux médullaires sont présents dans le système vasculaire du *Phytolacca dioica* (L.). Formé de deux à quatre brins vasculaires collatéraux, ayant leur pôle libérien orienté vers un centre commun, la structure primaire de ces faisceaux est composée. Un cambium est formé entre le xylème et le phloème des brins et s'étend pour encercler le phloème de tout le faisceau. Après une période d'activité cambiale, les faisceaux médullaires deviennent amphivasaux. Comme c'est le cas pour les espèces à phyllotaxie en hélice, le système vasculaire primaire est organisé en sympodiums. Les faisceaux médullaires forment les parties distales des traces foliaires médianes et continuent en position médullaire pour un nombre de noeuds égal au dénominateur de la fraction phyllotactique caractérisant une tige donnée. Lors du passage d'un faisceau médullaire dans une feuille, deux ou trois brins cribro-vasculaires se détachent du cylindre vasculaire pour former un nouveau faisceau médullaire. Le nombre de faisceaux médullaires dans une tige est ainsi maintenu. Des variations sur ce modèle sont présentes dans les régions basales des pousses juvéniles et dans les régions basales et apicales des pousses florifères adultes. Les auteurs discutent des relations entre la phyllotaxie et le passage des brins cribro-vasculaires dans la moelle et proposent une nouvelle classification des systèmes vasculaires à faisceaux médullaires.

## Article:

### INTRODUCTION

The Phytolaccaceae are a chiefly tropical and subtropical family of trees, shrubs, woody climbers, and herbs, some of whose members possess anomalous secondary thickening (Solereder 1908; Metcalfe and Chalk 1950, 1983). In addition to this anomalous thickening the vascular system of *Phytolacca dioica* includes medullary bundles. Solereder (1908, after earlier authors), Balfour and Philipson (1962), and Wheat (1977) have described these bundles as concentric, but a detailed investigation of these bundles has never been undertaken.

Despite continuing interest in the pattern of evolution of primary vascular systems (Beck et al. 1982) there are few studies of primary systems which include medullary bundles. Jones (1912) and Wilson (1924) are the only authors to provide relatively detailed reconstructions of primary vascular systems with medullary bundles. Puklawska (1972) partially reconstructs the primary vascular system of *Bougainvillea glabra* (Nyctaginaceae) and Balfour and Philipson (1962) provide a brief account of this system in *Phytolacca dioica*. Descriptions of nodal vascularization and medullary bundle structure without attempts to reconstruct the overall vascular pattern are more common (Dastur 1925; Maheshwari 1930; Inouye 1956; Davis 1961; Pant and Mehra 1961; Govil 1972; Pant and Bhatnagar 1975).

The aims of the present study on *P. dioica* are twofold: (i) to obtain exact knowledge of the anatomical structure of the medullary bundles; (ii) to obtain a detailed understanding of the organization of the primary vascular system and the relationship of the medullary bundles to this system. The latter will provide the basis for a study of the secondary body including the origin of the successive cambia. In addition to the above mentioned aims we will clarify some of the relationships between phyllotaxy and the direction of leaf trace divergence and will provide a brief review of the organization of the medullary bundle system in various species.

### MATERIALS AND METHODS

Shoots of *Phytolacca dioica* were collected from juvenile plants growing on the Givat Ram campus of The Hebrew University of Jerusalem. Adult as well as juvenile shoots were harvested from a large tree growing on the grounds of the School of Gardening and Landscaping in Petah Tiqwa. A voucher specimen from this tree was deposited in the Herbarium of The Hebrew University of Jerusalem (Laston 7-83-77-1).

Sections of both formaldehyde — alcohol — acetic acid fixed (90 mL 70% EtOH, 5 mL glacial acetic acid, 5 mL 40% formaldehyde) and fresh material were cut with a Reichert sliding microtome at 70, 100, and 200  $\mu$ m. The sections were then tied onto slides and stained. Young pieces of stem were dehydrated through a tertiary butyl alcohol series, embedded in paraffin, and sectioned with an American Optical model 820 rotary microtome at a thickness of 8-20  $\mu$ m, depending on the age of the tissue. The sections were mounted on slides with Bissing's modified Haupt's adhesive (Bissing 1974). Free-

TABLE 1. Phyllotactic patterns in *Phytolacca dioica*

Phyllotactic fraction	Direction of helix locally reversed	Number of shoots with sinistrorse helices	Number of shoots with dextrorse helices
3/8	No	13	6
3/8	Yes	2	1
2/5	No	1	—
2/5	Yes	—	—
2/7	No	1	—
2/7	Yes	—	1
Total		17	8
Irregular	—		1

hand sections were used to allow rapid investigation of variability in nodal and internodal structure. A total of 8 juvenile and 19 adult stems were examined. An additional juvenile stem was cleared for the study of the primary vascularization. The sections were stained with a mixture of safranin O and alcian green (Joel 1983), destained in ethanol, and either dehydrated, transferred to xylene and mounted in Cedrax (artificial Canada balsam), or rehydrated and mounted in glycerin jelly or Karo syrup (Johansen 1940). Freehand sections were mounted in 50% glycerin.

Clearings were made by soaking the material in lactic acid followed by 5% sodium hydroxide and hydrofluoric acid. The cortex was then carefully stripped away. After initial observation the vascular cylinder was stained with 1% basic fuchsin in an ammonium hydroxide solution (Boke 1970). Staining improved the distinctness of the vascular strands only slightly.

Connections between vascular strands determined by the above methods were confirmed, in part, by staining the xylem of the vascular bundles of a living shoot. The blade of a lower leaf was removed from a juvenile stem and a dye reservoir containing a dilute aqueous solution of methylene blue was attached to the cut petiole (Green 1925). After approximately 3 h the stem was harvested and the course to the dye was followed by sectioning with a Reiehert sliding microtome.

Phyllotactic fractions were determined by the external superposition of leaves along a stem and verified by an examination of the vascular system. A justification of this method is provided in Kirchoff (1984).

## RESULTS

### *Shoot types and phyllotaxy*

*Phytolacca dioica* is a South American dioecious tree (Fig. 1) of very rapid growth. Flowering specimens produce both adult and juvenile shoots. The former bear flowers in terminal racemes and are renewed by the growth of an axillary bud which produces a shoot similar in structure to the parent shoot. The juvenile shoots generally occur as root sprouts and continue vegetative growth for a whole season. They are much thicker than the flowering shoots and may reach a considerable length in one growing season. The largest juvenile stem observed in the present study was 1.1 m high and produced ca. 50 leaves.

The phyllotaxy of *P. dioica* is variable. All of the adult shoots examined have either 3/8 or irregular phyllotaxy, while the phyllotaxy of the juvenile shoots are 2/7, 2/5, 3/8, or irregular. Shoots with 2/7 phyllotaxy are characterized by a divergence angle of approximately 99.5°, while those with 2/5 or 3/8 phyllotaxy have divergence angles of approximately 137.5° (Dormer 1972). Two types of phyllotactic irregularities occur in the shoots studied. In a single juvenile shoot a 2/7 phyllotactic pattern changes abruptly to a more or less bijugate phyllotaxy. The more common phyllotactic irregularity is the reversal of the genetic helix in a local region of the stem. This usually involves only two leaves in any one region and appears to be caused by a reversal of the sequence of leaf initiation. It is often associated with an abnormally short internode between these leaves.

In *P. dioica*, the direction of rise of the phyllotactic helix may be either sinistrorse or dextrorse. A summary of the phyllotactic patterns observed in the shoots examined is presented in Table I. All of the terms and diagrams used here to describe and illustrate phyllotaxy are based on observations from the outside of the stem. The terms anodic (in the direction of the rise of the phyllotactic helix) and cathodic (in the direction opposite to the rise of the helix) refer to phyllotactic helices that rise from older to younger leaves.

### *Structure of the medullary bundles*

The medullary bundles of *P. dioica* have been described as concentric vascular bundles (Solereder 1908; Balfour and Philipson 1962; Wheat 1977). However, a developmental study reveals that they become so only through the activity of a vascular cambium. In their primary state the medullary bundles consist of two or three collateral vascular strands with their phloem poles oriented toward a common center (Fig. 2). A primary medullary bundle is, thus, a composite structure. In the following we will distinguish between the individual vascular strands and the medullary bundles composed of these strands.

As reported by Balfour and Philipson (1962), the vascular strands depart from the vascular cylinder to give rise to a medullary bundle which traverses a number of internodes and departs into a leaf to form the median vein. The pattern of departure from the vascular cylinder and the pattern of splittings and fusions of the vascular strands along the bundles may differ even in a single stem. However, some generalizations may be made. Two or three collateral vascular strands generally leave the vascular cylinder, enter the pith, and give rise to a medullary bundle (Figs. 3, 8). Just before leaving the cylinder all or some of the departing strands form connections with adjacent vascular strands which remain in the cylinder. When three strands enter the pith two of them fuse just above the point of entry so that only two strands can be distinguished in the medullary bundle immediately above this level (Fig. 2). As the medullary bundle attains a more central position in the pith, the two component vascular strands reorient so that their phloem poles face each other (compare Figs. 2 and 3). Splittings and fusions of the xylem and phloem groups occur, often independently, along the course of the medullary bundle.

Just below the insertion of a leaf, changes occur in the orientation of the vascular strands of the medullary bundle which enters the leaf. Three strands normally enter a leaf, the largest of which is centrally located and normally oriented, i.e., with the phloem toward the outside and xylem towards the center of the stem. The other two strands are much smaller, inverted, and are situated on the outer sides of the larger strand with their phloem poles facing the phloem of the larger

strand (Fig. 4). This difference in size between the vascular strands exists throughout the length of the medullary bundle and is most pronounced just below the node. It can already be observed at an early stage of medullary bundle differentiation when more vessel elements can be seen in one of the vascular strands than in the other(s) (Fig. 2).

The cambium of the medullary bundle first appears between the primary xylem and phloem groups of the component vascular strands and then extends laterally to form a complete ring. The presence of a functioning cambium is identified by the appearance of xylem fibers. With continuing cambial activity the medullary bundles achieve a more amphivasallike struc-

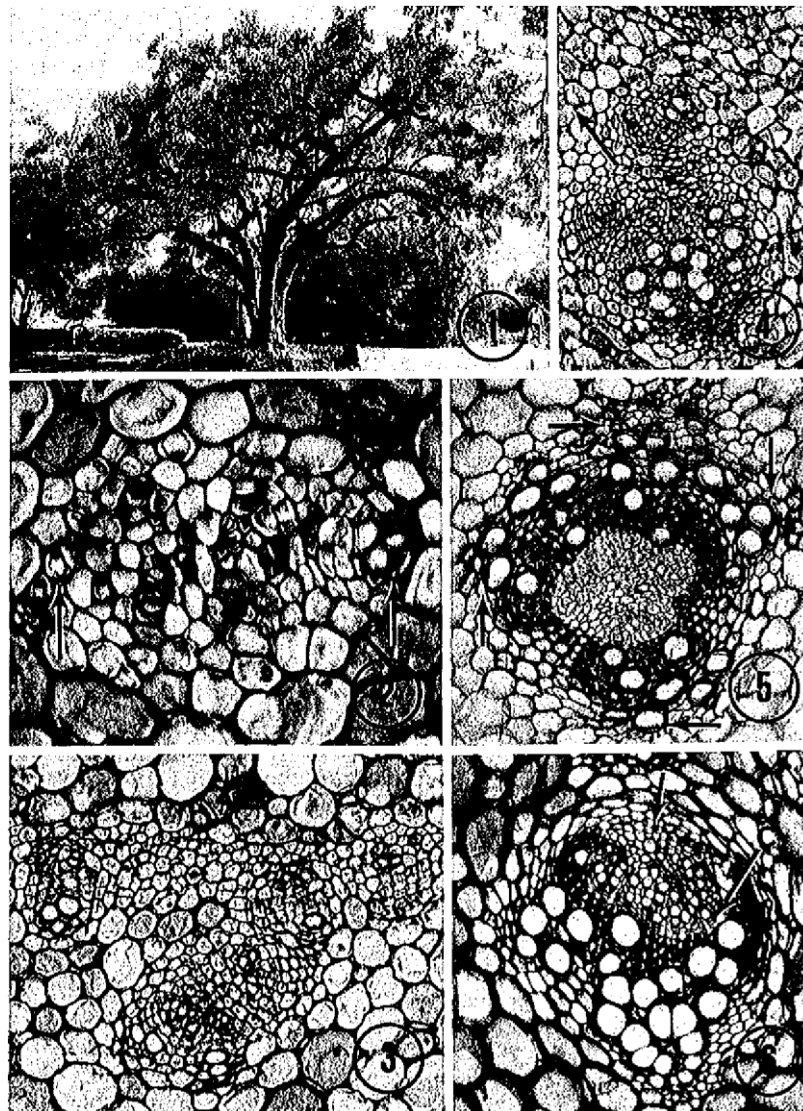


FIG. 1. A mature tree of *Phytolacca dioica* growing on the grounds of the School of Gardening and Landscaping in Petah Tiqwa, Israel. FIG. 2. Cross section of a young medullary bundle consisting of two vascular strands immediately above the level at which it departs from the vascular cylinder. The arrows indicate the protoxylem poles of the developing strands, X425. FIG. 3. Cross section showing the departure of two vascular strands from the vascular cylinder. x 128. FIG. 4. Cross section of a medullary bundle just below the insertion of the leaf to which this bundle departs. The arrow shows the direction in which the leaf base lies. x89. FIG. 5. Cross section of a medullary bundle after the beginning of cambial activity. The arrows indicate the primary xylem poles. x96. FIG. 6. Cross section of a medullary bundle with secondary growth just above its departure from the vascular cylinder. The arrow indicates the area of cambial discontinuity. C, cambium, X93.

ture. The secondary xylem fibers produced around the circumference are the main feature contributing to the concentric appearance of the medullary bundle. The production of secondary vessels is maximal next to the primary xylem groups and gives the whole bundle a lobed appearance (Fig. 5).

At the locations where vascular strands leave the vascular cylinder to form a medullary bundle and when a medullary bundle departs into a leaf, the cambium of the bundle becomes discontinuous between the component vascular strands. Where vascular strands leave the vascular cylinder, cambial connections do not develop between the strands and the cylinder but the cambium connecting the strands to each other is maintained (Fig. 6). As the strands reorient to form the medullary bundle, a connection is made between the free ends of the cambium to form the cambium of the medullary bundle. At the level at which a medullary bundle departs into a leaf, cambial connections among the component strands do not develop and the strands depart into the leaf independently. The component strands are normally oriented when leaving the cylinder and when departing into a leaf.

### *The pattern of the primary vascular system*

The organization of the primary vascular system of *P. dioica* is in accordance with the general principles of vascular organization described by Beck et al. (1982) and Kirchoff (1984). In a shoot with 2/5 phyllotaxy there are five sympodia and every second sympodium includes the median trace of the next leaf in the ontogenetic spiral (Fig. 7). Since the medullary bundles of *P. dioica* represent the distal parts of the median leaf traces (Fig. 7), there are five medullary bundles and alternate medullary bundles depart to successive leaves (Figs. 8, 10). Analogous situations are found in shoots with 3/8 and 2/7 phyllotaxy. In these cases there are eight and seven sympodia and medullary bundles, respectively, and every third or second

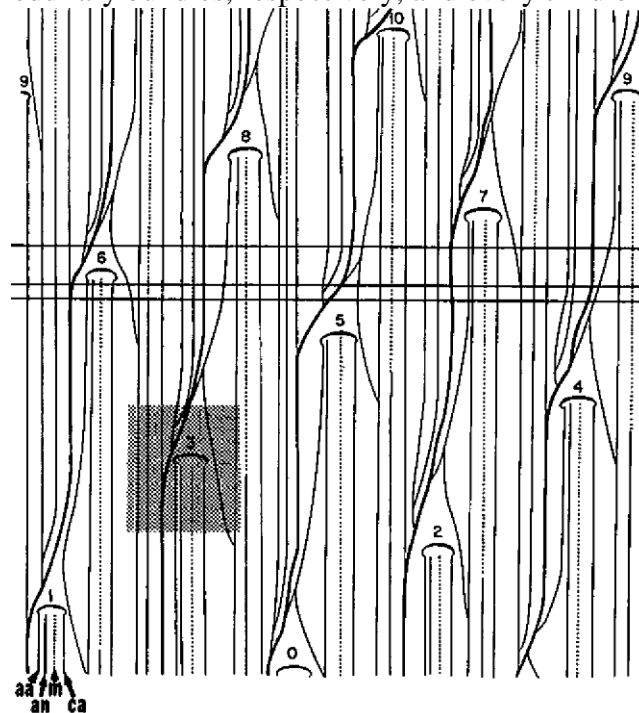


FIG. 7. Simplified diagram of the primary vascular system of a shoot of *P. dioica* with 2/5 sinistrorse phyllotaxis. One line represents several vascular strands. Five sympodial bundles are shown by heavy lines. The leaf traces and their branches are represented by thin lines. The courses of the medullary bundles are marked by dotted lines.

At the point where solid and dotted lines meet, several vascular strands move into the pith to form the medullary bundle. Horizontal lines *a*, *b*, and *c* indicate the levels of the sections shown in Fig. 8. The shaded box indicates a region of the stem comparable to the one enlarged in Fig. 9. *an*, anodic leaf trace; *ca*, cathodic leaf trace; *m*, median leaf trace (medullary bundle); *aa*, additional anodic leaf trace.

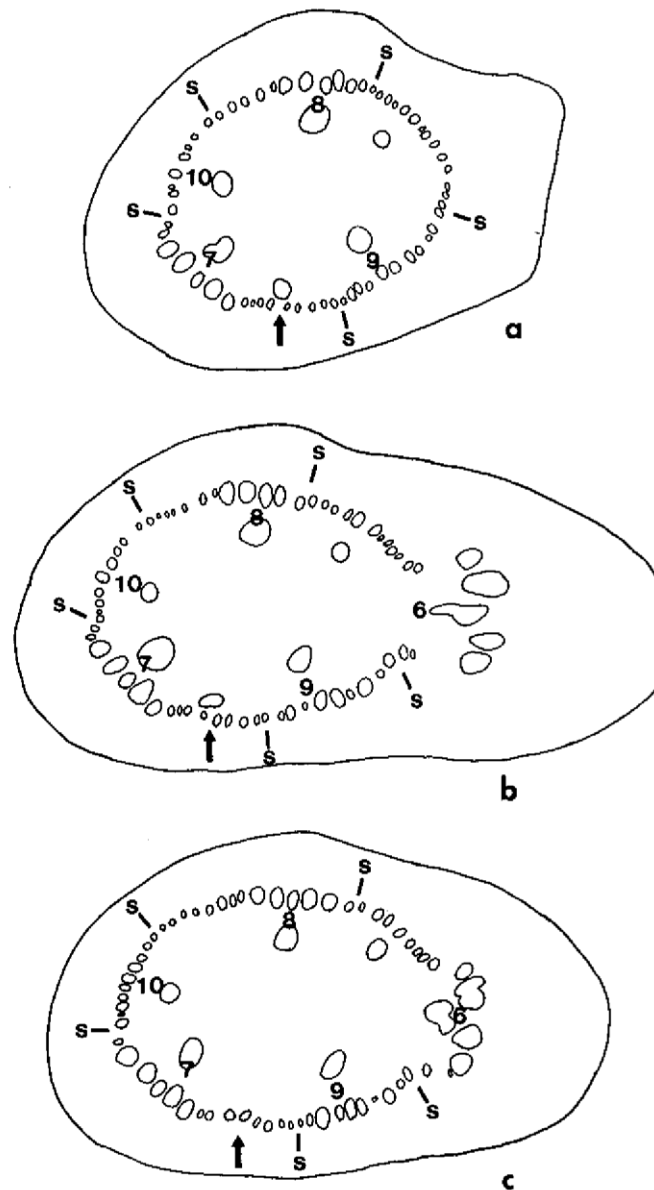


FIG. 8. Tracings of sections at the levels indicated in Fig. 7. The numbers indicate the vascular supply to the leaves with the same numbers as in Fig. 7. *s*, regions the sympodial bundles; arrow, region in which vascular strands depart the vascular cylinder to give rise to a medullary bundle.

sympodium and medullary bundle vascularizes a sequentially formed leaf.

As has been found previously (Beck et al. 1982; Kirchoff 1984) the number of internodes between the branchings of a sympodial bundle (the major vascular bundle of a sympodium which continues without interruption and gives rise to the leaf traces) that give rise to median leaf traces is generally equal to the denominator of the phyllotactic fraction. The length of the median as



well as of the lateral leaf traces was investigated only in shoots with  $2/5$  phyllotaxy (Fig. 7). After branching from a sympodial bundle the median (Fig. 7, trace m) and anodic (Fig. 7, trace an) lateral traces traverse just over 12 nodes before departing into a leaf. The cathodic leaf traces (Fig. 7, trace ca) traverse eight nodes before entering a leaf. An additional anodic (Fig. 7, trace aa) lateral trace branches from the sympodial bundle three nodes below the anodic trace and departs into the leaf.

The number of nodes traversed by the medullary bundles is directly related to the phyllotactic fraction of a shoot. In shoots with  $2/5$  phyllotaxy the medullary bundles traverse five nodes, in shoots with  $3/8$  and  $2/7$  phyllotaxy they traverse eight and seven nodes, respectively. This fact is a corollary of the number of sympodia in the stem and of the fact that when a medullary bundle enters a leaf a new medullary bundle, arising from the same sympodium as the old one, departs from the vascular cylinder into the pith (Figs. 7, 8, 10). A constant number of medullary bundles is, thus, maintained in the stem.

The level at which vascular strands enter the pith to form a new medullary bundle varies between shoots and, occasionally, within the same shoot. Vascular strands may depart from the vascular cylinder into the pith up to a node and a half above or below the point where the adjacent medullary bundle, which belongs to the same sympodium, enters a leaf. However, it is more common for the changes in position of both bundles to occur at the same level.

The vascular strands which enter the pith and form new medullary bundles may do so either on the cathodic or anodic side of the medullary bundle departing to the leaf. In shoots with  $2/5$  and  $2/7$  phyllotaxy the new medullary bundle arises on the cathodic side (Figs. 8, 10). In shoots with  $3/8$  phyllotaxy it arises on the anodic side. These relationships hold regardless of the direction of the ontogenetic helix (sinistrorse or dextrorse). They can be understood in relation to leaf ar-



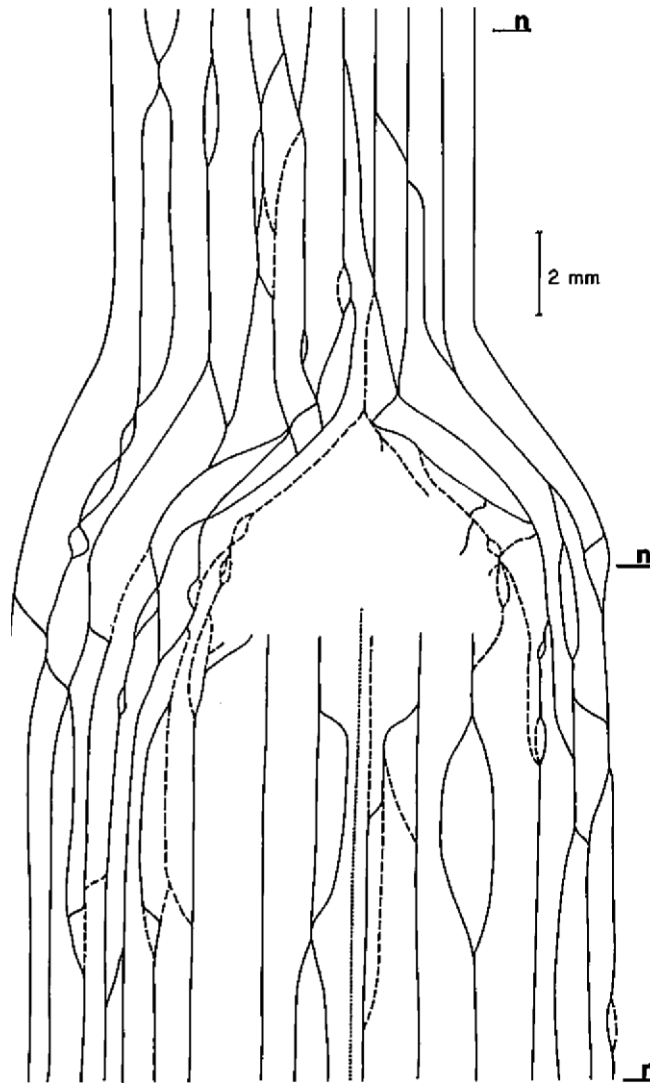


FIG. 9. Diagram of the course of the vascular strands in a region of a mature stem. This enlargement is comparable to the shaded region in Fig. 7. Each line represents one vascular strand. Only the longitudinal dimension is to scale. *n*, node; —, vascular strands with both primary and secondary xylem and phloem; ---, vascular strands with only secondary xylem and phloem; • • medullary bundle.

range at the apex (Fig. 11) and are dealt with more fully in the discussion.

Up to this point only the primary vascular pattern in the region close to the shoot apex has been dealt with (Fig. 7). The fully developed primary vascular system is much more complex (Fig. 9). Two factors contribute to this complexity. First, as the shoot matures numerous additional primary bundles differentiate in the vascular cylinder. Second, when secondary growth begins numerous vascular bundles containing only secondary xylem and phloem are formed between the existing primary bundles (Figs. 9, 12). These secondary bundles are especially common in the regions of the vascular cylinder adjacent to departing leaf traces (Fig. 9). By the time the first supernumerary cambium is initiated, the initial vascular cylinder is packed with bundles and the pattern of the primary vascular system is obscured. Connections between primary and secondary vascular bundles occur throughout the vascular cylinder (Fig. 9).

### *Variability of the vascular pattern*

A number of variations are found in the primary vascular pattern of *P. dioica*. In the most common variant the distal portions of the medullary bundles do not enter the leaves directly from the pith, but join the vascular cylinder at some distance below the leaf and only depart into the leaf after traversing a number of internodes. The total number of internodes traversed by these bundles, first in the pith and then in the vascular cylinder, remains equal to the denominator of the phyllotactic fraction. The distal portion of a medullary bundle, which is incorporated in the vascular cylinder, is positioned slightly centripetally in relation to the neighboring lateral traces of the same leaf (Fig. 14). After secondary growth begins, cambial continuity is established between the medullary bundle and the lateral leaf traces in this region (Fig. 14).

The phenomenon of a medullary bundle joining the vascular cylinder before entering a leaf does not occur at random. Within a shoot it is most commonly found at the lower nodes, i.e., at those produced soon after budbreak. In juvenile shoots the frequency of this phenomenon decreases gradually as growth continues until all of the medullary bundles depart directly into the leaves without first joining the vascular cylinder. As a result the number of medullary bundles in the pith varies from one level to another, the smaller number generally occurring at the base of the shoot. There is, however, considerable variation in the number of medullary bundles in the lowest nodes. In most shoots no medullary bundles were found in the lowest internode, but in a few, one collateral bundle was found in the pith and in a single shoot four such bundles were found. The upper region of a juvenile shoot has the regular vascular pattern described in the previous section.

In flowering shoots, which generally have  $3/8$  phyllotaxy, a larger proportion of the medullary bundles rejoin the vascular cylinder before departing into a leaf. In these shoots it is common to find a gradual increase in the number of medullary bundles from zero, to three, four, or five in the middle of the stem and then a decrease to one or zero below the inflorescence. The lack of sufficient flowering material has made it impossible to relate this decrease to the vascularization of the inflorescence. A further difficulty arises from the existence of flowering shoots in which no decrease in the number of medullary bundles occurs below the inflorescence.

An additional anomaly is found in one juvenile shoot. In this shoot a vascular strand, the xylem of which is of secondary origin only, enters the pith from the vascular cylinder one to several nodes below the entry of the vascular strands which form the regular medullary bundle. As the irregular bundle continues up the stem, the amount of xylem and phloem is gradually reduced until only small parenchyma cells, which resemble primary xylem parenchyma, remain. At approximately this level the remnant of the irregular strand and the primary xylem pole of the regular medullary bundle fuse. The departure of the median trace into the leaf is normal.

Variations in the organization of the primary vascular system may also be associated with phyllotactic anomalies. In the simplest case the genetic helix is reversed in a local region of the stem apparently because of a reversal of the sequence of leaf initiation. This reversal usually involves only two leaves and is accompanied by a short internode. Since the position of two leaves is exchanged there is a one node extension of the traces to the leaf now initiated higher and a truncation of the traces to the lower leaf.

The last type of variation observed is in nodal structure. The

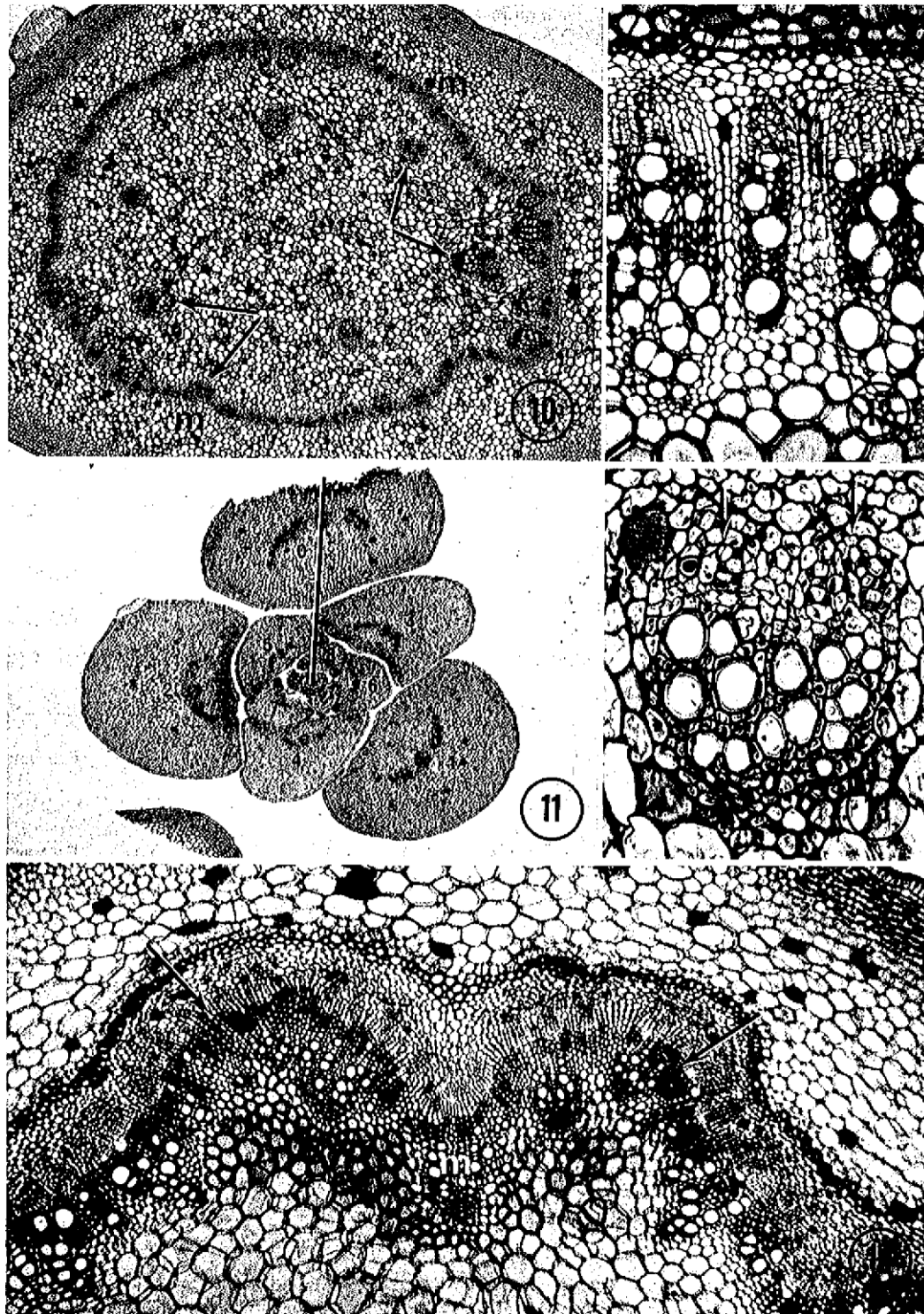


FIG. 10. Cross section of a young stem with  $2/5$  sinistrorse phyllotaxis showing the vascular cylinder and medullary bundles of five sympodia. The pairs of arrows indicate medullary bundles belonging to the same sympodium. The formation of a new medullary bundle (*m*) takes place on the cathodic side of the departing leaf vascular supply. x26. FIG. 11. Cross section through the apex of a shoot with  $3/8$  sinistrorse phyllotaxis. The leaves are numbered from older to younger beginning with an arbitrarily chosen leaf (leaf 0). See text for explanation. x 23. FIG. 12. Cross section of a vascular strand consisting solely of secondary xylem and phloem

between two strands containing both primary and secondary tissues. x 108. Flo. 13. Cross section of a leaf trace showing two phloem groups (arrows) outside a single xylem group. x282. FIG. 14. Cross section of a mature stem showing the incorporation of a medullary bundle (*m*) into the vascular cylinder below its departure to a leaf. The arrows show the limits of the leaf vascular supply. x60.

number of traces supplying a leaf, excluding the medullary bundle, is variable. At the gross level of observation between three and six lateral traces are found. More detailed observation reveals that the number of primary xylem groups varies between five and eight, and the number of primary phloem groups varies between 8 and 18. The number of phloem groups is greater than the number of xylem groups because two or more phloem groups are often present opposite one xylem

TABLE 2. Data on medullary bundle structure

Species and family	Primary bundle type	Cambium present in medullary bundle	Bundle type after secondary growth	Medullary bundles vascularize		Source
				Leaves	Buds	
Part I. Only leaf traces occupy a medullary position						
<i>Bougainvillea glabra</i> (Nyctaginaceae)	Collateral	Yes	—	Yes	No	Puławska 1965
<i>Phytolacca dioica</i> (Phytolaccaceae)	Composite	Yes	Amphivasal	Yes	No	This study
Part IIa. Leaf traces and sympodial bundles occupy a medullary position, leaf traces depart directly to the leaf						
<i>Boerhavia diffusa</i> (Nyctaginaceae)	Collateral	Yes	—	Yes	Yes	Maheshwari 1930; Pant and Mehra 1961
<i>Mirabilis jalapa</i> (Nyctaginaceae)	Collateral	—	—	Yes	—	Inouye 1956
<i>Dahlia lehmanni</i> (Compositae)	Collateral, inverted collateral	No	—	Yes	Yes	Davis 1961
<i>Amaranthus graecisans</i> (Amaranthaceae)	Collateral	—	—	Yes	Yes	Wilson 1924
<i>Amaranthus hybridus</i> (Amaranthaceae)	Collateral	—	—	Yes	—	Wilson 1924
Part IIb. Leaf traces and sympodial bundles occupy a medullary position; leaf traces join the vascular cylinder before departing to the leaf						
<i>Dianthera americana</i> (Acanthaceae)	Collateral	Yes	Amphicribal	Yes	Yes	Jones 1912
<i>Argyrea nervosa</i> (Convulvulaceae)	Phloem only, collateral, bicollateral, inverted collateral	Yes	Bicollateral, amphicribal, biamphicribal	Yes	Yes	Pant and Bhatnagar 1975
<i>Argyrea roxburghii</i> (Convulvulaceae)	Inverted collateral	Yes	Amphicribal	Yes	Yes	Pant and Bhatnagar 1975
<i>Achyranthes aspera</i> (Amaranthaceae)	Collateral, xylem on two sides of the phloem	—	—	Yes	Yes	Dastur 1925; Joshi 1934
Part III. Medullary bundles do not contribute to vascularization of leaves"						
<i>Chenopodium album</i> (Chenopodiaceae)	Collateral	—	—	No	—	Wilson 1924
<i>Chenopodium ambrosioides</i> var. <i>anthelminticum</i> (Chenopodiaceae)	Collateral	—	—	No	—	Wilson 1924
<i>Beta cycla</i> (Chenopodiaceae)	Collateral	—	—	No	—	Wilson 1924
<i>Celosia cristate</i> (Amaranthaceae)	Collateral	—	—	No	—	Wilson 1924

<sup>a</sup>See text for discussion.

group (Fig. 13). This difference appears to be a result of the independent anastomosing of phloem and xylem strands.

## DISCUSSION

A large number of families have been reported to possess medullary bundles (Solereder 1908; Metcalfe and Chalk 1950, 1983). While a comprehensive survey is beyond the scope of this paper some general conclusions may be drawn from the more recent literature (Table 2).

### *Structure of the bundles*

According to the literature medullary bundles are generally collateral and normally oriented. However, *Dahlia lehmanni*, *Argyreia nervosa*, and *Argyreia roxburghii* have at least some inversely oriented bundles and some individuals of *Achyranthes aspera* are reported to have medullary bundles with xylem on two sides of the phloem. It is unclear whether these latter bundles are of primary origin or if they are partially secondary because neither Dastur (1925) nor Joshi (1934) report on the presence of secondary growth. The medullary bundles of *P. dioica* are compound bundles. Each is composed of from two to four collateral vascular strands the phloem poles of which are directed towards a common center.

In the few species in which secondary growth has been studied there is a tendency for the medullary bundles to become concentric (amphivasal or amphicribal; Table 2). Secondary growth generally starts in a collateral bundle and these bundles most often become amphicribal after a period of cambial activity. When a functional cambium differentiates in a bicollateral (biphloic) medullary bundle of *Argyreia* (Convolvulaceae), the bundle develops an unusual structure termed biamphicribal by Pant and Bhatnagar (1975). *Phytolacca dioica* is the only species whose medullary bundles become amphivasal as a result of secondary growth.

### *The structure and organization of the medullary bundles*

In all of the species reviewed for this paper the medullary bundles were found to be part of the leaf trace systems and, in most cases, with the exception of *Bougainvillea glabra* and *P. dioica*, to contribute to the vascularization of the buds.

On the basis of De Bary's (1877, cited and discussed in Pant and Bhatnagar 1975) classification of plants with medullary bundles and of a detailed analysis of the vascular systems reported in the more recent literature we propose a new classification based on the placement of the medullary bundles and the organization of the primary vascular system. We recognize two types of vascular systems with medullary bundles. Type 1 (Table 2, part I): vascular systems with only leaf traces occupying a medullary position. The sympodial bundles and all the minor bundles are part of the vascular cylinder. Type 2 (Table 2, part II): vascular systems with both leaf traces and sympodial bundles in a medullary position.

The often incomplete descriptions of the vascular system of these latter species make it difficult to propose subdivisions of type 2. However, two general patterns of leaf trace departure may be recognized: (i) (Table 2, part Ha) the direct departure of the main leaf traces from the pith to the leaf; (ii) (Table 2, part IIb) the leaf traces occupying a medullary position first join the vascular cylinder, traverse one or several internodes, and only then enter a leaf.

In addition to the species included in the above classification, Wilson (1924) describes the vasculature of some species of the Chenopodiaceae and Amaranthaceae in which the medullary bundles do not pass directly into leaves (Table 2, part III). In these species the sympodial bundles, or parts of them, occupy a medullary position. These sympodial bundles do not occupy as central a position in the pith as do the medullary bundles of the other species reviewed. In some cases they are situated only one or two cell layers more internally than the other bundles of the vascular cylinder. This type of arrangement of bundles is typical of the Chenopodiaceae

where it is common to find ribbed stems with vascular bundles arranged in slightly irregular rings. Some of the bundles are, thus, situated slightly more internally than others. These facts cause us to question the application of the term medullary to such bundles. It seems more appropriate to restrict the term medullary to bundles which are distinctly located in the pith and at least some of which enter the leaves.

#### *The relationship between leaf arrangement and the movement of vascular strands into the pith*

The vascular strands which enter the pith and form new medullary bundles may do so either on the cathodic or the anodic side of the medullary bundle departing to the leaf. The side of the leaf on which the new medullary bundle arises is related to the phyllotactic fraction of the stem but not to the direction of the ontogenetic helix. Shoots with  $2/5$  or  $2/7$  phyllotaxy always give rise to the new medullary bundles on the cathodic side of the leaf trace, while in shoots with  $3/8$  phyllotaxy they arise on the anodic side. This relationship is identical to that found for the direction in which the median leaf traces branch form sympodial bundles. When the phyllotactic fraction is  $1/3$ ,  $3/8$ ,  $8/21$ , or  $3/11$ , the branching is anodic; when the fraction is  $2/5$ ,  $5/13$ ,  $2/7$ , or  $5/18$ , it is cathodic (Namboodiri and Beck 1968).

These relationships can be understood by considering the relationship between the arrangement of leaves at the apex and the vascular pattern. Figure 11 is a cross section of an apex with a divergence angle of approximately  $137.5^\circ$ . The leaves are numbered along the ontogenetic helix from older to younger. A line drawn from the center of the apex through the center of an older leaf (leaf 0) passes closer to the centers of leaves 3, 5, 8, 13, 21, etc. (i.e., leaves numbered in the Fibonacci series), then to the centers of any other leaves. The centers of the higher numbered leaves are closer to the line than are those of the lower numbered leaves. It can also be seen that leaves numbered alternately in the Fibonacci series lie on alternate sides of the line. The mathematical basis of these facts has been explained by Richards (1951), Mitchison (1977), and Jean (1984).

Now, if the phyllotactic fraction characterizing this apex is  $p/n$ , then every  $n$ th leaf will be linked to the same sympodium. However, since there are no leaves that arise directly above one another, the leaves will be linked either in the cathodic (left side of the line in Fig. 11) or anodic (right side of the line in Fig. 11) direction from the previous leaf. When  $n = 3, 8, 21$ , etc., the trace linkage is anodic; when  $n = 5, 13$ , etc., it is cathodic.

Similar arguments can be made for apices which have divergence angles other than  $137.5^\circ$ . All of the divergence angles found in plants with helical phyllotaxy (Dormer 1972) show the same properties as this angle. The direction of the median trace linkage is, thus, determined by the divergence angle and the denominator of the phyllotactic fraction and is independent of the direction (sinistorse or dextrorse) of the phyllotactic helix. As is well known, the denominator is also the number of sympodia in a stem and is related to leaf arrangement at apex in a precise way (Kirchoff 1984).

Namboodiri and Beck (1968) give a similar argument to the one presented above to explain the direction of trace divergence, but curiously, Beck et al. (1982) fail to point out this relationship. For this reason and because there has been a lack of attention to the relationships between leaf arrangement and vascular pattern we have decided to include an explanation of trace divergence



here. It is hoped that this contribution along with that of Kirchoff (1984) will stimulate a renewed interest in this feature of plant construction.

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